## Assessment of Vegetation Stress Using Reflectance or Fluorescence Measurements

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#### **ABSTRACT**

Current methods for large-scale vegetation monitoring rely on multispectral remote sensing, which has serious limitation for the detection of vegetation stress. To contribute to the establishment of a generalized spectral approach for vegetation stress detection, this study compares the ability of high-spectral-resolution reflectance (R) and fluorescence (F) foliar measurements to detect vegetation changes associated with common environmental factors affecting plant growth and productivity. To obtain a spectral dataset from a broad range of species and stress conditions, plant material from three experiments was examined, including (i) corn, nitrogen (N) deficiency/excess; (ii) soybean, elevated carbon dioxide, and ozone levels; and (iii) red maple, augmented ultraviolet irradiation. Fluorescence and R spectra (400-800 nm) were measured on the same foliar samples in conjunction with photosynthetic pigments, carbon, and N content. For separation of a wide range of treatment levels, hyperspectral (5-10 nm) R indices were superior compared with F or broadband R indices, with the derivative parameters providing optimal results. For the detection of changes in vegetation physiology, hyperspectral indices can provide a significant improvement over broadband indices. The relationship of treatment levels to R was linear, whereas that to F was curvilinear. Using reflectance measurements, it was not possible to identify the unstressed vegetation condition, which was accomplished in all three experiments using F indices. Largescale monitoring of vegetation condition and the detection of vegetation stress could be improved by using hyperspectral R and F information, a possible strategy for future remote sensing missions.

Since the turn of the 20th century, anthropogenic impacts have significantly altered the natural environment. The current biomass production in agriculture and commercial forestry is achieved primarily by applying nitrogen (N)-enriched fertilizers and raising soil N levels. Tropospheric ozone (O<sub>3</sub>) and carbon dioxide (CO<sub>2</sub>) levels have also increased, as has ultraviolet (UV) radiation reaching the ground level, all of which lead to progressive alteration in natural and cultivated ecosystems (U.S. Climate Change Science Program, 2005). Accurate remote sensing (RS) monitoring, regularly providing synoptic views of the dynamic ecosystem parameters, is required for vegetation monitoring, timely stress detection, understanding the direction of environmental change, and making effective management decisions. To contribute toward the establishment of an

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accurate, generalized RS approach for vegetation monitoring, this investigation comparatively evaluates the capabilities of spectral reflectance (R) and fluorescence (F) measurements for the detection of vegetation stress associated with common environmental factors, such as N deficiency and elevated  $CO_2$ ,  $O_3$ , and UV.

# Nitrogen, Carbon Dioxide, Tropospheric Ozone, and Ultraviolet Effects on Vegetation Physiology

Plant carbon (C) sequestration and biomass production are driven by N availability because N is involved in photochemical processes and is one of the primary resources regulating plant growth. Corn, a C<sub>4</sub> species, has higher requirement for N than most other plants or crop species (Meisinger, 1984). However, excessive N applications may suppress crop yield and biomass allocation, and N runoff from agricultural and urban areas can cause soil acidification and leaching of essential minerals (e.g., cations P<sup>2+</sup>, K<sup>+</sup>, Mg<sup>2+</sup>, and Ca<sup>2+</sup>) (McMurtrey et al., 1994). Because most terrestrial ecosystems evolved in N-limited environments, careful monitoring is needed to understand species and ecosystems dynamics in an N-rich world.

Globally, industry and transportation are adding almost 7 trillion kg yr<sup>-1</sup> of C to the atmosphere, and atmospheric CO<sub>2</sub> concentrations are on the rise (U.S. Climate Change Science Program, 2005). Although this enrichment of the atmosphere portends hazards, it offers the potential benefit to increase plant production. At elevated CO<sub>2</sub>, the photosynthetic efficiency and yield of important crops such as corn, soybean, cotton, peanut, rice, wheat, sorghum, and oats may experience increase (i.e., C<sub>3</sub> plants more than C<sub>4</sub>) (Heagle et al., 1998; Tiedemann and Firsching, 2000). Under natural conditions, limiting factors (e.g., water and N deficits and weather extremes) and the resulting changes to plant physiology would mitigate the positive effects (Tuba, 2005).

Due to anthropogenic activities,  $O_3$  pollution is on the rise in the troposphere, presenting a threat to agricultural crops and human health (U.S. Climate Change Science Program, 2005). Because of its reactive nature, tropospheric  $O_3$  is the primary component of air pollution that causes negative affects on vegetation physiology, growth, and yield (Krupa and Kickert, 1989; Mulchi et al., 1995; Krupa et al., 2001; Kim et al., 2003). During the day, leaf stomata are normally open, permitting the entry of  $CO_2$  for photosynthesis, which enables  $O_3$  to enter the stomata (Reich, 1987; Larcher, 1995). Ozone

**Abbreviations:** Car, carotenoids; Chl, chlorophyll; ChlF, chlorophyll fluorescence; cps, counts per second; D, derivative; F, fluorescence; K, potassium; LS means, least square means; NDVI, normalized difference vegetation index; PRI, photochemical reflectance index; R, reflectance; REIPw, wavelength position of the red edge inflection point; RS, remote sensing; SLM, specific leaf mass; TM (1–7), spectral bands on LandsatTM; USDA, United States Department of Agriculture; UV, ultraviolet.

can cause a range of adverse effects, including premature leaf aging, early onset of senescence, growth and yield reductions, and altered sensitivity to biotic and abiotic stresses (Skärby et al., 1993; Krupa et al., 2001; Fuhrer, 2003; Jäger et al., 2003). Plants have evolved protective mechanisms to repair  $O_3$  injury before damage is severe, such as increased production of the antioxidant vitamins C and E, polyamines, and specialized enzymes (Melhorn and Wellburn, 1987). Therefore, careful monitoring and timely detection of  $O_3$  damage may enable management decisions that limit the adverse effects.

Numerous investigations addressing the combined effect of CO<sub>2</sub> and O<sub>3</sub> on plants' physiologic responses indicate that the positive effects of CO<sub>2</sub> on vegetation growth are negated, partially or completely, by elevated O<sub>3</sub> concentrations (Krupa and Kickert, 1989; Leblanc, 1998; Kim et al., 2003). Extended foliar exposure to O<sub>3</sub> typically reduces CO<sub>2</sub> fixation, and further inhibition of CO<sub>2</sub> uptake occurs through O<sub>3</sub>-induced reduction in chloroplast function, smaller stomatal opening, or CO<sub>2</sub> loss through respiration (Guzy and Heath, 1993; Jäger et al., 2003). On the other hand, the negative effects of O<sub>3</sub> stress may be partially or fully ameliorated under increased atmospheric CO<sub>2</sub> concentrations (McKee et al., 2000; Kim et al., 2003). However, in the agricultural areas of the midwestern USA, tropospheric O<sub>3</sub> is increasing more rapidly than CO<sub>2</sub>. The corn-soybean agricultural complex is the largest ecosystem of the contiguous USA and is one of the most important for USA exports and international food security. Systematic monitoring of this system would provide significant economic benefits (U.S. Climate Change Science Program, 2005).

Since the 1970s, human activities have disrupted the natural balance between stratospheric O<sub>3</sub> synthesis and breakdown, resulting in chemical depletion of the O<sub>3</sub> layer and an increase of UV-B levels reaching the Earth's surface (especially in the Southern hemisphere). The effects of UV-B radiation on vegetation are mostly damaging, often resulting in toxic or mutagenic DNA products that inhibit plant function and depress biomass allocation (Middleton and Teramura, 1993; Krupa et al., 2001). Common protective responses of field-grown plants to elevated UV-B radiation are the increased synthesis of UV-absorbing compounds in the foliage and the development of thicker leaves as a result of an increase in the thickness of the upper epidermis layer and the spongy parenchyma (Sullivan et al., 2002). Because many of the UV-B-induced changes in physiology and function are linked to reductions in plant growth and productivity, elevated UV-B levels can be considered a significant environmental stress factor for agricultural and natural ecosystems (Middleton and Teramura, 1993; Tegelberg et al., 2001; Sullivan et al., 2003).

### **Spectral Detection of Vegetation Stress**

As changes in an ecosystem occur, alterations in foliar chemistry and membrane structure result in changes in vegetation spectral signatures (Rock et al., 1986, 1994; Martin, 1994; Martin and Aber, 1997; Entcheva et al., 1999, 2004); thus, spectral observations could offer a sensitive, physiologically based indicator of vegetation stress. Current methods for monitoring vegetation bio-

physical and growth parameters rely on broadband (≥20 nm bands) satellite data, such as those on Landsat Thematic Maper, MODIS (Moderate Resolution Imaging Spectroradiometer), and SPOT (Systeme Pour l'Observation de la Terre) satellites. The commonly used visible and near-infrared (700–1200 nm) R broadband vegetation indices are primarily associated with foliar chlorophyll (Chl) levels at the top of the canopy, not providing information about the function of the canopy as a whole. Therefore, their use has serious limitations for early detection of vegetation stress or for estimation of vegetation biophysical parameters not directly related to Chl content. As compared with broadband, high-spectral-resolution (>10 nm) R parameters are much better correlated with the amount of foliar Chl (Carter, 1994; Carter et al., 1995; Carter and Spiering, 2000; Carter and Knapp, 2001) and with the amount of projected green leaf area of canopies and landscapes observed by RS platforms (Walter-Shea et al., 1992; Entcheva et al., 1999; Carter and Spiering, 2000; Entcheva, 2000). Only the PRI index (PRI1 = [R530 - R570]/[R530 + R570]) has been associated with vegetation photosynthetic activity (Gamon et al., 1997). Recent technological developments, such as the launch of the first Earth Observing satellite by NASA, are making space-borne hyperspectral data more widely available. High-spectral-resolution R data can provide a significant improvement over the broadband indices for the detection of changes in vegetation condition and for monitoring vegetation decline (Entcheva, 2000; Entcheva et al., 2004). However, R measurements are not able to directly assess vegetation function.

### **CONCLUSIONS**

Although R and F variables were successful in detecting vegetation stress responses in each of the three experiments, only a few of the R and F variables performed rigorously across all three experiments, including the D715/D705 R ratio and the green/far-red (F530/ F740) F ratio. This study established that high-spectralresolution reflectance (5-10 nm) can provide a significant improvement over the currently used broadband indices for the detection of changes in vegetation physiologic condition. Narrow-band R indices, especially those using first derivatives, performed best and were found strongly correlated to pigment, C, and N contents and to the C/N ratio. The narrow band R indices successfully discriminated among treatment responses, but they could not identify the unstressed (optimal) plant condition within a stress gradient, as was demonstrated for the F (F530/F740) data. Consequently, F and R data provided complimentary information.

The current RS methods for assessment of photosynthetic function and C sequestration, based on the combined satellite observations and modeling efforts, could be improved by incorporating the physiologic information obtained using hyperspectral R and F observations. More research is needed to extend our findings to other vegetation types and species and to evaluate the association of photosynthetic function to F emissions produced from visible excitation wavelengths (e.g., 400–650 nm) and high-spectral-resolution R properties.